I and II outer, the IB4/ret population terminates deeper in a narrow band within lamina II inner in association with a layer of neurons expressing high levels of protein kinase C γ (PKCγ). A functional correlation has also been made between the two types of sensory neurons. Loss of the PKCγ gene using homologous recombination in mice prevents the development of neuropathic pain following partial nerve section (Malmberg et al., 1997), while experimental inflammation of the hind paw in rats and mice results in an increased expression of substance P in the peptide/trkA neurons. This has led to the suggestion that chronic inflammatory pain is mediated largely by the peptide/trkA-containing sensory neurons that terminate in superficial laminae, while neuropathic pain resulting from peripheral nerve damage is mediated by the IB4/ret population that terminates in the deeper regions of lamina II (Snider and McMahon, 1998).

To determine which sensory neurons express the VR1 protein, antibodies were generated to the predicted carboxyl terminus of VR1, and an immunohistochemical analysis was performed. The results are surprising. Staining of the sensory ganglion revealed that about 80% of both IB4/ret- and peptide/trkA-containing sensory neurons express VR1 protein-like immunoreactivity. In other words, there appears to be a small but substantial population of sensory neurons that do not express the VR1 but which previously have been shown to be sensitive to capsaicin. This data, along with binding studies with tritiated resiniferatoxin, suggests that there are additional vanilloid receptors.

The immunohistochemical analysis also revealed an unexpected heterogeneity in the IB4/ret population of sensory neurons that terminates in the inner portion of lamina II. It has previously been shown that the medial and lateral regions of the dorsal horn of the spinal cord represent distal and proximal parts of the hindlimb, respectively (Devor and Claman, 1980). What is unique in the present report is that whereas the IB4/ret population that terminates in the medial aspect of lamina II inner shows colocalization with VR1, the IB4/ret population that terminates in the lateral aspect of lamina II inner shows virtually no colocalization with VR1. This data suggests that either the IB4/ret populations which innervate the distal aspects of a limb express the VR1, whereas those that express the proximal aspect of the limb do not, or that there is differential transport of the VR1 receptor protein to the spinal cord in these two populations of sensory neurons. There is a precedent for differential transport of a receptor in primary afferents: the neuropeptide Y Y1 receptor that is expressed in sensory neurons is found in the cell bodies but rarely in the axons that terminate in the spinal cord (Zhang et al., 1994). Also, Robert Elde's lab has suggested that the synthesis and trafficking of a receptor and neurotransmitter can be coregulated in sensory neurons. Thus, disruption of the preprotachykinin gene inhibits the translation of δ opiate receptor—a protein normally coexisting in the same synaptic vesicles as substance P (Dray and Rang, 1998). Thus, the expression of VR1 could potentially be regulated at the transcriptional, translational, or axoplasmic transport level. Differential regulation of transport would result in modified terminal sensitivity within discrete populations of sensory neurons.

Why do pain researchers find this work so interesting? The majority of therapies used today for the treatment of chronic pain (opiates, aspirin, and codeine) have been utilized for over a century and have significant side effects, especially with long-term use. One of the most promising avenues for discovering new molecules to treat chronic pain is to understand the molecular and cellular mechanisms that underlie transducer function in primary afferent nociceptors. The beauty of the present work is that it begins to provide a cellular and molecular framework for understanding the vast and often provocative literature on the biological actions of capsaicin. It would be highly surprising if future work on capsaicin and VR1 does not contribute significantly to our knowledge of sensory transducer function and ultimately to the development of new therapies for treating persistent pain.

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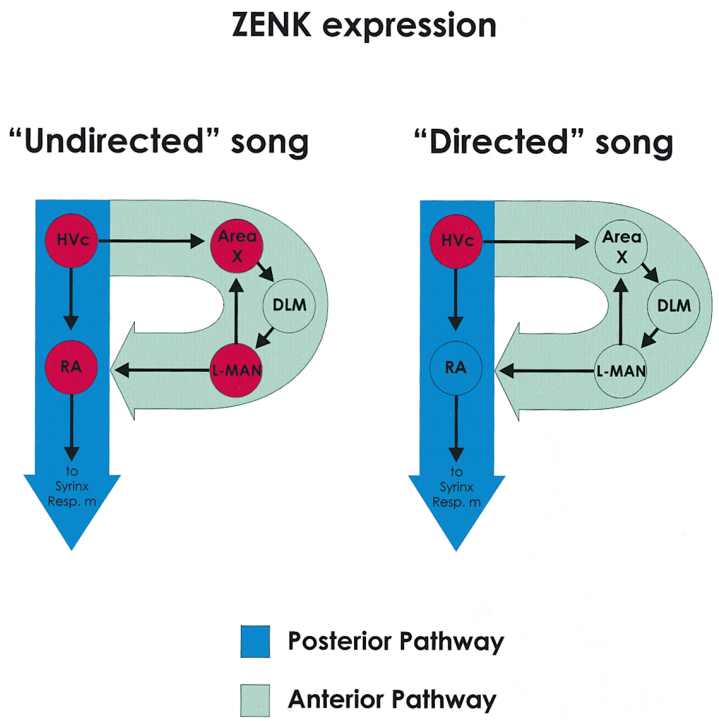
Modulation by Social Context Sheds New Light on Mechanisms of Vocal Production

Molecular tools are used increasingly to probe aspects of nervous system function. In many cases, such studies



Schematic of ZENK Expression Patterns during Production of the Same Song in Two Different Social Contexts

High levels of ZENK expression (shown in red) are observed in RA as well as area X and /MAN of the "indirect" anterior pathway when male zebra finches sing alone or in the presence of other males ("undirected" song). Singing in the presence of a female ("directed" song) does not induce ZENK expression in these nuclei. The forebrain nucleus HVc, also known as high vocal center, shows high levels of ZENK expression during both directed and undirected song. Expression patterns in the thalamic nucleus DLM do not appear to be correlated with singing.

are performed without a solid understanding of the behavioral relevance of the specific molecules, synapses, or circuits under investigation. The potential danger of ignoring behavioral context is that the significance of certain results may go unnoticed. Given the sometimes subtle variations of an animal's behavioral repertoire, relating specific molecular or physiological events to the correct behavioral context may be difficult unless a detailed understanding and analysis of the animal's behavior is performed. This point is nicely illustrated by Jarvis et al. (1998) in this issue of *Neuron*. Studying song production in zebra finches, they combine careful behavioral analysis with gene expression to show that a remarkably subtle variation in behavior (whether the bird sings alone or toward a female) can lead to striking differences in gene expression.

The male zebra finch only produces one song, which consists of a variable number of introductory notes followed by several repetitions of a stereotyped sequence of syllables. The song is produced when the bird is alone ("undirected" song) or in the presence of a female ("directed" song), and, although containing the same song elements, directed song is often delivered slightly faster, contains a few more introductory notes, and is accompanied by a courtship dance. The structure of each song type, however, is nearly indistinguishable, even when viewed on a sound spectrograph. The brain structures responsible for the production of these different song types are organized as an interconnected network of nuclei known collectively as the song system. This system can roughly be divided into a "direct" vocal motor pathway, referred to by Jarvis et al. (1998) as the posterior pathway, and an "indirect" vocal pathway known as the anterior pathway (see figure). Whereas the posterior pathway is known to be hierarchically organized and conveys song motor commands to the syrinx (avian vocal organ) and various respiratory muscles (Margoliash, 1997; Suthers, 1997), the functional significance of the anterior pathway is less well understood. Previously thought to be involved exclusively during the phase of song acquisition in juvenile birds, recent studies now implicate the anterior pathway in song maintenance in adult birds as well (Doupe and Solis, 1997). The recent findings by Jarvis et al. (1998) suggest that this pathway may also play an important role in modulating song motor output within the behavioral, or social, context in which song is produced.

By carefully monitoring different song types and controlling for the overall number of songs produced within a given experimental period, expression of the immediate early-gene ZENK, as shown by in situ hybridization, is shown to increase 10- to 40-fold in most nuclei of the song system during undirected song. Remarkably, with the notable exception of nucleus HVc, these nuclei show little or no expression during directed song (see figure). Because ZENK is though to be an indicator of neural activation (Chaudhuri, 1997), these results suggest that activity patterns in RA and the anterior pathway differ considerably depending on the context in which singing occurs. The authors conclude that the differences in ZENK expression are likely caused by singing-related motor activity, since expression patterns are similar to those observed in deafened birds. Because of the recent findings, however, that auditory flow into the song system may be dependent on behavioral context (Schmidt and Konishi, 1998), the possibility nevertheless remains that differential "gating" of auditory feedback signals may contribute to the different patterns of ZENK expression.

While the differential expression of ZENK during directed and undirected singing provides new insight into the control of vocal production, the behavioral significance of these different context-dependent song types,

as well as why brain areas are differentially activated during these behaviors, is not clear. One possibility is that the differential activation of the anterior pathway during undirected and directed song may serve to coordinate other motor behaviors that are generally associated with these different song types. For example, when the male directs his song to a female, the anterior pathway may well be optimized to coordinate, or bind, song motor activity with other motor behaviors such as the courtship dance, which is only observed during directed singing.

The use of immediate-early genes, such as ZENK, as markers for neural activity, provides a powerful tool that can be used in identifying new ways in which neural circuits may be organized. As the authors point out, however, interpreting the relationship between ZENK expression and neural activity should be done cautiously. A case in point is the pattern of ZENK expression observed in nucleus RA. This structure, which forms part of the direct motor pathway, is known to be active during singing (Margoliash et al., 1997), exhibits high levels of c-*fos* expression (Kimpo and Doupe, 1997), and yet expresses relatively low levels of ZENK. Clearly, the findings by Jarvis et al. (1998) set the stage for a detailed characterization of this phenomenon at the electrophysiological level. Although technically challenging, recording neural activity in birds singing under different behavioral contexts is now an active area of research and promises to yield exciting new findings.

Increasing anatomical and neurochemical evidence (Bottjer and Johnson, 1997; Luo and Perkel, 1998; Jarvis et al., 1998) suggests that there are significant parallels between the anterior song control pathway (HVc → area X → DLM → *l*MAN) and the mammalian cortical–basal ganglia pathway (cerebral cortex → basal ganglia → thalamus → cortex). There are functional parallels as well: the basal ganglia, in addition to being involved in motor execution, are also thought to be involved in motor learning, in sensorimotor integration, and in the control of motor acts that require motivational or cognitive drive (Graybiel et al., 1994). Of specific interest in the context of the results obtained by Jarvis et al. (1998) is the finding that "motor" neural activity in striatum (to which area X may be homologous) can be modulated by behavioral context. A striking example of such context-dependent neural activation can be shown in striatal input neurons during a memory-guided saccade task (Kawagoe et al., 1998). By combining saccade tasks with a differential reward schedule, the authors show that firing patterns that are normally predictive for a given spatial location can become significantly depressed depending on whether that saccade target is, or is not, paired with a reward. Thus, although the motor act, the saccade, is identical in both cases, pairing a motor act with a reward (a motivational, or emotional, drive) can completely alter neural responses within the striatum.

As in the mammalian striatum, area X is strongly innervated by midbrain dopaminergic areas (Bottjer and Johnson, 1995). These inputs in mammals are thought to convey reward-related and behavioral context–dependent information to the striatum (Graybiel et al., 1994). Given the similarities between the two systems, a detailed characterization of dopaminergic inputs into the song system is likely to yield important insights into the mechanisms underlying the differences observed between directed and undirected singing. More generally, important mechanistic insight into the functioning of the song system is likely to emerge from a deeper understanding of the mammalian cortical–basal ganglia pathway and its role in motor control and function, while, conversely, work on the mammalian basal ganglia will likely benefit from a deeper understanding of the song system.

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Strange Bedfellows? Protein Degradation and Neurological Dysfunction

Angelman syndrome (AS) is a severe neurological disorder characterized by seizures, ataxia, and cognitive dysfunction. AS occurs in ~1 of every 15,000 births and appears to arise from the functional loss of a maternally expressed imprinted gene or genes. The majority of cases are attributable to a chromosomal deletion on the maternally inherited chromosome 15 (see figure). Loss of the functional maternal allele in AS also occurs by paternal uniparental disomy (UPD), in which two copies of chromosome 15 are inherited from the father, by defects in the imprinting process (Nicholls et al., 1998), and it recently has been shown to arise from gene mutation of the maternally inherited allele of *UBE3A*, an E3 ubiquitin ligase (see figure; Malzac et al., 1998). The paper by Jiang et al. (1998) in this issue of *Neuron* confirms the importance of UBE3A in AS, by demonstrating in a mouse model that a mutation in the maternal allele of *Ube3a* is sufficient for an AS-like phenotype. This result raises the interesting question of why a mutation in a gene involved in a general cellular process such